# Biology and Management of Almond Scab and Alternaria Leaf Spot

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## A. Summary

Scab caused by *Fusicladium carpophilum* (currently classified as *Venturia carpophila*) and Alternaria leaf spot caused by *Alternaria alternata* and *A. arborescens* are economically important summer diseases of almond, especially in locations with high humidity and poor air circulation. For scab, dry environments during bloom and early stages of fruit development (petal fall period) tend to reduce the disease, whereas wet springs are favorable. For Alternaria leaf spot, warm day-time environments, cool nights, and frequently scheduled irrigations to keep up with evapotranspiration of trees provide favorable conditions for dew formation and consequently disease. *Alternaria* spp. are ubiquitous air-borne plant pathogens and typically cause disease in high-production orchards that are planted densely and have high water and nitrogen inputs. The result is low air movement and consistent wetness periods from dew. Many of the current pollinators are highly susceptible to Alternaria leaf spot (e.g., Monterey, Carmel).

We continued our evaluations of the pathogens from different areas in the state. *Venturia* and *Alternaria* spp. only reproduce asexually by conidia in California. Based on molecular diversity analyses, however, two species of *Venturia* may exist sympatrically (co-existing) and occupy the same niche on almond tissues and may even be present in the same lesions. The two populations were designated based on their slow (S-) or fast (F-) growth rate in culture. Both types of isolates had an optimum temperature of 25C for spore germination 15 h after plating onto potato dextrose agar. Drying spores at 24% RH at 25C for 1 to 7 days on glass slides reduced germination (viability) from 49.4% at 0 days to 12.3, 6.0, 8.1, and 4.7% for 3, 4, 5, and 7 days for S-isolates, and from 98% to 71.8%, 68.4% and 34% for F-isolates in the same time period.

In dormant fungicide trials for managing scab on almond, FRAC codes (FC) M3 (ziram), M4 (captan), and M5 (chlorothalonil) were applied in 3% oil mixtures without any phytotoxicity. Dormant treatments with M3 or M4 (both moderately effective) or M5 (highly effective) fungicides delay scab inoculum production on overwintering twig lesions and are an anti-resistance strategy (i.e., a smaller population is exposed to selection processes) that align in-season, springtime treatments for scab, Alternaria leaf spot, and rust to May and June timings.

Field trials to evaluate in-season applications of organic and conventional fungicides were established in early May for Alternaria leaf spot and scab in each year of the project. Emphasis was on mixtures of different FCs to prevent selection of resistance in target populations. Resistance has been detected in *Alternaria* and *Venturia* spp. to FC 7 and 11 fungicides. Boscalid, fluopyram, fluxapyroxad, isofetamid, and penthiopyrad are registered, and pydiflumetofen, pyraziflumid, fluindapyr, and the experimental GWN10570 are in development. We collected diseased almond leaves and evaluated isolates of *Alternaria* spp. for their sensitivity to selected SDHI (FC 7) fungicides and established that certain mutations in DNA sequences of SDH subunits B, C, and D correspond with resistance to selected SDHI sub-

groups. The highest incidence of resistance occurred against boscalid, fluxapyroxad, penthiopyrad, and pyraziflumid. Only moderate resistance ($EC_{50}$ <0.5 ppm) has been detected to fluopyram and isofetamid and low resistance to the new pydiflumetofen. We identified cross resistance patterns among SDHI fungicides that correlated with mutations SDHB-H277Y, SDHB-277L, or SDHC-H134R (most common). Resistance to newer SDHI sub-groups was detected before commercial introduction. Thus, variants most likely pre-exist, and non-detrimental mutations occur without significant fitness penalties to the pathogen.

DMI fungicides (FC 3) including the new Cevya are highly active against *Alternaria* isolates and against isolates molecularly identified as *V. carpophila*. A relatively narrow range of sensitivity was observed in baseline studies with DMI fungicides. Outliers were detected in laboratory studies; however, these fungicides remain highly effective in managing both diseases in the field.

In our in-season field trials on Alternaria leaf spot and scab, Regev, Cevya, the experimentals V-10424 and GWN-10570, as well as the pre-mixtures Miravis Top (=Miravis Duo), Miravis Prime, and UC-2 were evaluated and compared to other mixtures (e.g., Fervent, Fontelis + Teb, Luna Experience, and Quadris Top). In general, premixtures containing FC 3 were the most effective for scab and Alternaria leaf spot management. The trials also included organic compounds such as natural products (e.g., plant extracts, SARs), food grade treatments exempt from tolerance (e.g., potassium sorbate/sodium lauryl sulfate), experimental biocontrols (e.g., *Bacillus amyloliquifaciens*) and numbered compounds. Overall, these products performed poorly, and disease levels were not significantly different from the controls.

## B. Objectives

**1)** Determine population composition of the scab pathogen *V. carpophila* and occurrence of sexual reproduction in *Alternaria alternata* and the scab pathogen. ***Significant findings:*** *Neither species showed molecular evidence of sexual reproduction and both are only reproducing asexually in California. Two co-existing (sympatric) populations were identified for* V. carpophila *and were designated based on their slow (S-) or fast (F-) growth rate in culture. S- and F-isolates had optimum temperatures of 25 C for spore germination. Spore germination of S-isolates was dramatically reduced at 25 C, 24% RH to <5% after 7 days, whereas the F-isolates maintained relatively high germination (34%) even after 5 to 7 days under these conditions.*

**2)** For scab management, evaluate the effect of dormant treatments on development of spring sporulation. ***Significant findings:*** *Dormant treatments with M3 and M4 (both moderately effective) and M5 (highly effective) fungicides delay scab inoculum production on overwintering twig lesions and are an anti-resistance strategy.*

**3)** Evaluate new and registered fungicides for their efficacy as in-season applications against scab and Alternaria leaf spot. ***Significant findings:*** *Identified highly effective, in-season pre-mixtures or tank mixtures of FC 3+19, 3/7, 3/11, and 3/12. Rotational programs were identified to prevent selection of resistance. Biologicals and natural products were significantly less effective or ineffective against these diseases.*

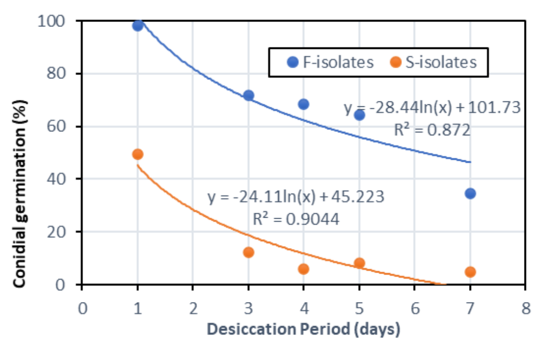
**4)** Establish baseline sensitivities, monitor for fungicide sensitivity shifts in the pathogens, and characterize mechanisms for resistance against SDHI and DMI fungicides. ***Significant findings:*** *SDHI and DMI baselines were established for several fungicides in FC 3 and 7. Sensitivities to SDHIs within each species varied over a wide range; less variability occurred to DMI fungicides. The highest incidence of resistance (>40 ppm) occurred against boscalid, fluxapyroxad, penthiopyrad, and pyraziflumid. Only reduced* *sensitivity ($EC_{50}$ <0.5 ppm) was detected to fluopyram and isofetamid; and most isolates were highly sensitive to the new pydiflumetofen. In molecular studies, cross resistance patterns among SDHI fungicides correlated with mutations SDHB-H277Y, SDHB-277L, or SDHC-H134R (most common). Resistance to SDHI sub-groups were detected before commercial introduction and variants most likely pre-exist.*

## C. Results and Discussion

1. **Determine population composition of the scab pathogen *V. carpophila* and occurrence of sexual reproduction in *A. alternata* and the scab pathogen.** *Alternaria* and *Venturia* spp. appear to only reproduce asexually by conidia in California. Still, evaluation of genotyping data using 32 microsatellite primers demonstrated high genetic diversity among 39 isolates of *V. carpophila* from almond, peach, and pecan, and isolates from almond and pecan were highly variable. Further genotyping using RAPD and UP-PCR differentiated distinct populations for each of these hosts and within the almond isolates. Cultural growth studies over a range of temperatures from 10 C to 35 C indicated that there were two populations with dissimilar growth rates at their optimum temperature of 25 C. Isolates were designated as slow (S-) or fast (F-) growing strains. Thus, based on molecular analyses and cultural growth rates, two populations of *V. carpophila* (or possibly two *Venturia* species) were identified from almond. These two populations co-exist sympatrically and occupy the same niche on almond leaves, fruit, and green shoots and may even be present in the same lesions.

Effects of wetness and drying on spore germination of S- and F-isolates demonstrated that conidia of the two populations responded differently to desiccation. Initial germination was 98% the F-isolates and 49.4% for S-isolates on Day 1. After air-drying and incubating at 25 C, 24% RH, for selected times, conidial germination generally decreased after 3, 4, 5, and 7 days (Fig. 1). Data were regressed after ln-transformation, and $R^2$ values were 0.87 and 0.90 for the F- and S-isolates, respectively. S-isolates dropped to <5% germination at the last day of sampling, whereas germination of F-isolates was 34%.

Fig. 1. Effect of desiccation on conidial germination of slow- (S-) and fast (F-) growing isolates representing two populations of *V. carpophila*. Initial germination percentage is shown on Day 1. Conidia were air-dried at 25 C, 24% RH, for selected times, re-hydrated, and placed onto PDA. Spore germination was determined after 18-24 h. Data were regressed using a ln-transformation.




**2. For scab management, evaluate the effect of dormant treatments on development of spring sporulation.** In dormant fungicide trials for managing scab on almond, the FCs M3 (ziram), M4 (captan), and M5 (chlorothalonil) were applied in 3% oil mixtures in January without any phytotoxicity. Dormant treatments with M3 and M4 (both moderately effective) and M5 (highly effective) fungicides significantly delayed scab inoculum production on overwintering twig lesions. This application timing has minimal impact on pollinators and is an anti-resistance strategy (i.e., a smaller population is exposed to selection processes) for fungicides that are subsequently applied because it delays and reduces sporulation of the scab pathogen and aligns in-season, springtime treatments for scab, Alternaria leaf spot, and rust to May and June. In-season applications combined with dormant treatments were the most effective programs in reducing incidence and severity of the disease on fruit (Table 1). Furthermore, the 5-wk- after petal fall timing has a minimal impact on pollinators.

**Table 1. Efficacy of fungicide treatments for management of scab of Winters almond - Sutter Co.**

| | | | Applications | | Dis. Incid. on fruit** | | Lesions on fruit | |
|---|---|---|---|---|---|---|---|---|
| | | | 1-30-20 | 4-14-20 | | | | |
| No. | Treatment* | Rate/A | Dormant | 5 wk after | (%) | LSD^ | No. | LSD |
| 1 | Control | --- | --- | --- | 73.6 | a | 8.3 | a |
| 2 | Bravo | 64 fl oz | @ | --- | 17.9 | cd | 0.5 | b |
| 3 | Bravo | 64 fl oz | @ | @ | 8.7 | d | 0.2 | b |
| 4 | Ziram | 96 oz | @ | --- | 54.6 | ab | 2.2 | b |
| 5 | Ziram | 96 oz | @ | @ | 20.3 | cd | 0.8 | b |
| 6 | Ziram + Inspire | 96 oz + 6 fl oz | --- | @ | 52.5 | b | 3.6 | b |
| 7 | Regev | 6 fl oz | --- | @ | 32.8 | bc | 1.2 | b |

* - Treatments were applied using an air-blast sprayer at a rate of 100 gal/A. Dormant treatments were done in combination with 3% oil.

**Table 2. Efficacy of fungicide treatments for management of almond scab – A. Colusa Co. cv. Carmel and B. Yolo Co. cv. Monterey.**

| A. cv. Carmel | | | | | Applications | | | Dis. Incid. on fruit | | Lesions on fruit | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| No. | Program | Treatment* | Rate/A | Adjuvant | 5-5 | 5-27 | 6-18 | (%) | LSD | No. | LSD |
| 1 | --- | Control | --- | --- | --- | --- | --- | 65.1 | a | 5.3 | a |
| 2 | Biologicals | Serifel | 8 oz | DyneAmic | @ | @ | @ | 59.4 | a | 3.5 | abc |
| 3 | | All Phase | 18 oz | DyneAmic | @ | @ | @ | 53.9 | a | 3.2 | abcd |
| 4 | | GWN 10474 | 28 oz | DyneAmic | @ | @ | @ | 56.7 | a | 3.4 | abc |
| 5 | | GWN 10474 | 35 oz | DyneAmic | @ | @ | 20 oz | 65.3 | a | 5.4 | a |
| 6 | Single | Regev | 6 fl oz | --- | @ | @ | @ | 23.9 | bc | 0.8 | de |
| 7 | | Cevya | 5 fl oz | DyneAmic | @ | @ | @ | 47.6 | ab | 2.5 | bcde |
| 8 | Mixture | Merivon + Serifel | 6.5 fl oz + 8 oz | DyneAmic | @ | @ | @ | 58.2 | a | 4.3 | ab |
| 9 | Pre- | Luna Experience | 8 + 8 fl oz | DyneAmic | @ | @ | @ | 8.9 | c | 0.2 | e |
| 10 | mixtures | Luna Sensation | 8 fl oz | DyneAmic | @ | @ | @ | 46.0 | ab | 2.4 | bcde |
| 11 | | Merivon | 6.5 fl oz | DyneAmic | @ | @ | @ | 44.3 | ab | 1.4 | cde |
| 12 | | UC-2 | 6 + 3.84 fl oz | DyneAmic | @ | @ | @ | 9.7 | c | 0.3 | e |
| 13 | Rotation | Ph-D + Teb | 6.2 + 8 oz | DyneAmic | @ | --- | @ | 15.0 | c | 0.5 | e |
| | | Quadris Top | 14 fl oz | DyneAmic | --- | @ | --- | | | | |

| B. cv. Monterey | | | | | Application | | | Dis. Incid. on fruit** | | Lesions on fruit | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| No. | | Treatment | Rate | Adjuvant | 5-13 | 6-3 | 6-24 | (%) | LSD^ | No. | LSD |
| 1 | --- | Control | --- | --- | --- | --- | --- | 93.1 | a | 14.0 | a |
| 2 | Biologicals | EcoSwing | 32 fl oz | DyneAmic | @ | @ | @ | 72.4 | ab | 9.7 | ab |
| 3 | | Timorex Act | 25 fl oz | DyneAmic | @ | @ | @ | 69.6 | abc | 9.6 | ab |
| 4 | | Serifel | 8 oz | DyneAmic | @ | @ | @ | 66.9 | bcd | 10.2 | ab |
| 5 | | All Phase | 18 oz | DyneAmic | @ | @ | @ | 56.1 | bcde | 8.6 | b |
| 6 | | LifeGuard | 4.5 oz | DyneAmic | @ | @ | @ | 67.2 | bcd | 8.6 | b |
| 7 | Rotation | LifeGuard | 4.5 oz | DyneAmic | @ | --- | --- | 70.8 | abc | 9.9 | ab |
| | | Double Nickel | 32 fl oz | DyneAmic | --- | @ | @ | | | | |
| 8 | Single | Regev | 6 fl oz | --- | @ | @ | @ | 38.0 | cdef | 3.2 | c |
| 9 | Mixture | Merivon + Serifel | 6.5 fl oz + 8 oz | DyneAmic | @ | @ | @ | 36.3 | def | 3.0 | c |
| 10 | Pre-mixtures | Quadris Top | 14 + 8 fl oz | DyneAmic | @ | @ | @ | 23.0 | ef | 2.1 | c |
| 11 | | EXP-AD | 13.7 fl oz | DyneAmic | @ | @ | @ | 11.5 | fg | 0.7 | c |
| 12 | | EXP-AF | 9.1 fl oz | DyneAmic | @ | @ | @ | 2.4 | g | 0.0 | c |

* - Treatments were applied using an air-blast sprayer at a rate of 100 gal/A. DyneAmic was used at 8 fl oz/100 gal (except for the Regev treatment).

**3. Evaluate new and registered fungicides for their efficacy as in-season applications against scab and Alternaria leaf spot.** Field trials to evaluate in-season applications of organic and conventional fungicides were established in early May for Alternaria leaf spot and scab in each year of the project. Emphasis was placed on mixtures of different FRAC Codes (FCs) to prevent selection of resistance in target populations. Resistance has been detected in *Alternaria* and *Venturia* spp. to FC 7 and 11 fungicides. For scab, most biologicals evaluated were ineffective or inconsistent (e.g., Serifel), whereas single and pre-mixtures of conventional fungicides had a significantly lower incidence of disease as compared to the untreated control on both Carmel and Monterey cultivars (**Table 2**). Among the conventional materials, pre-mixtures (e.g., FC 3/7, 3/11, experimentals UC-2, Miravis Top, Miravis Prime) and tank mixtures (e.g., 3+19) were the most effective. In evaluations of disease severity, among biologicals only LifeGuard and All Phase significantly reduced the disease on cv. Monterey but not on cv. Carmel as compared to the control. Still, the conventional fungicides significantly reduced the disease most dramatically from an average of 14 lesions per fruit to <3 or to fractions of a lesion.

Table 3. Efficacy of fungicide treatments for managing Alternaria leaf spot of cv. Monterey - Yolo Co.

| | | | | Applications | | | Incidence** | | Severity | | Defoliation | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| No. | Program | Treatment* | Rate (/A) | 5-13 | 6-3 | 6-24 | % | LSD^ | Rating | LSD | Rating | LSD |
| 1 | --- | Control | — | — | — | --- | 100.0 | a | 3.1 | a | 2.8 | a |
| 2 | Biological | Serifel | 8 oz | @ | @ | @ | 90.2 | abc | 1.8 | bc | 1.6 | bc |
| 3 | Single | Fontelis | 20 fl oz | @ | @ | @ | 86.0 | abc | 1.6 | bc | 1.7 | b |
| 4 | | Quash liquid | 3 fl oz | @ | @ | @ | 65.7 | bc | 1.2 | bc | 1.0 | bcd |
| 5 | | Cevya | 5 fl oz | @ | @ | @ | 65.1 | c | 1.0 | c | 0.9 | cd |
| 6 | | Magister | 32 fl oz | @ | @ | @ | 91.0 | abc | 2.0 | b | 1.7 | b |
| 7 | | GWN 10570 | 10 fl oz | @ | @ | @ | 72.0 | bc | 1.6 | bc | 1.6 | bc |
| 8 | Mixtures | Merivon + Serifel | 6.5 fl oz + 8 oz | @ | @ | @ | 72.4 | bc | 1.5 | bc | 1.2 | bcd |
| 9 | | Quash + Velum | 3 + 6 fl oz | @ | @ | @ | 77.3 | bc | 1.1 | c | 0.8 | d |
| 10 | | Fontelis + Teb | 20 fl oz + 8 oz | @ | @ | @ | 84.1 | abc | 1.2 | bc | 0.9 | bcd |
| 11 | Pre-mixtures | Luna Experience | 8 fl oz | @ | @ | @ | 76.8 | abc | 1.3 | bc | 1.1 | bcd |
| 12 | | Luna Sensation | 8 fl oz | @ | @ | @ | 95.5 | ab | 1.8 | bc | 1.3 | bcd |
| 13 | | Merivon | 6.5 fl oz | @ | @ | @ | 88.1 | abc | 1.8 | bc | 1.2 | bcd |
| 14 | | UC-2 | 7 fl oz | @ | @ | @ | 73.0 | bc | 1.2 | bc | 0.6 | d |
| 15 | Rotations | Cevya | 5 fl oz | @ | — | --- | 74.2 | bc | 1.0 | c | 0.6 | d |
| | | Merivon | 6.5 fl oz | — | @ | | | | | | | |
| | | Ph-D | 6.2 oz | — | — | @ | | | | | | |
| 16 | | Fontelis | 10 fl oz | @ | — | --- | 71.0 | bc | 1.0 | c | 0.8 | d |
| | | Quash liquid | 3 fl oz | — | @ | | | | | | | |
| | | Ph-D | 6.2 oz | — | — | @ | | | | | | |

* - Treatments were applied using an air-blast sprayer at a rate of 100 gal/A, all in combination with DynAmic (8 fl oz/A).

** - For evaluation of disease, a severity scale was used on 25-35 leaves of each single-tree replication: 0 (healthy), 1 (<33%), 2 (33-66%), and 4 (>66%) of leaf surface diseased. Defoliation ratings were: 0 = no defoliation, 1 = <25%, 2 = 26-50%, 3 = 51-75%, 4 = >75%.

For Alternaria leaf spot, Serifel, Magister, Fontelis, Fontelis+Teb, Luna Experience, and Merivon were ineffective in reducing the incidence of disease in this trial, whereas most conventional fungicides had a significantly lower incidence as compared to the untreated control with 100% incidence on cv. Monterey in Yolo Co. (**Table 3**). Resistance to FC 7 fungicides at this location is likely responsible for these results. All treatments, however, significantly reduced disease severity to $\leq$1.8 from that of the control with 3.1 lesions per fruit and improved tree health by reducing defoliation to a rating of $\leq$1.7. The best treatments had ratings of <1 as compared to the control with a 2.8 rating.

In a second trial on cv. Monterey in Colusa Co., disease was lower in the untreated control with 65.5% incidence. Only the Rhyme treatment was ineffective, and this was due to the leaf phytotoxicity observed with three consecutive applications of the fungicide. All treatments were conventional fungicides, and they significantly reduced disease incidence (**Table 4**). Under lower disease pressure, the severity of disease on fruit was also reduced to a rating of $\leq$0.58 as compared to 1.12 for the untreated control. Thus, fungicide treatments and programs are more effective under low disease pressure emphasizing integrated cultural practices and fungicide management programs. Moreover, fungicide programs are effectively shifting the disease (i.e., as indicated by severity and defoliation ratings) in high-production orchards (i.e., high density and high inputs of water and nitrogen) to later into the fall season where defoliation will occur naturally on the deciduous almond tree.

Table 4. Efficacy of fungicide treatments for managing Alternaria leaf spot of cv. Monterey - Colusa Co.

| No. | Program | Treatment* | Rate (/A) | Applications 5-16 | Applications 6-14 | Applications 8-21 | Incidence % | Incidence LSD | Severity Rating | Severity LSD^ |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | --- | Control | --- | --- | --- | --- | 65.5 | ab | 1.12 | a |
| 2 | Single | Rhyme*** | 7 fl oz | @ | @ | @ | 86.7 | a | 1.36 | a |
| 3 | | Pyraziflumid | 4.7 fl oz | @ | @ | @ | 36.4 | cde | 0.44 | bcd |
| 4 | | Fontelis | 20 fl oz | @ | @ | @ | 42.5 | bcd | 0.58 | b |
| 5 | | Ph-D | 6.2 oz | @ | @ | @ | 19.8 | de | 0.20 | cd |
| 6 | | UC-1 | 5 fl oz | @ | @ | @ | 26.6 | cde | 0.29 | bcd |
| 7 | Mixtures | Quash + Intuity | 2 oz + 2 fl oz | @ | @ | @ | 25.2 | cde | 0.29 | bcd |
| 8 | | Fontelis + Teb | 20 fl oz + 8 oz | @ | @ | @ | 23.7 | cde | 0.26 | bcd |
| 9 | Pre-mixtures | Luna Experience | 8 fl oz | @ | @ | @ | 20.4 | de | 0.23 | bcd |
| 10 | | Quadris Top | 14 fl oz | @ | @ | @ | 13.5 | e | 0.14 | d |
| 11 | | Merivon | 6.5 fl oz | @ | @ | @ | 45.6 | bc | 0.54 | bc |
| 12 | | UC-2 | 7 fl oz | @ | @ | @ | 26.1 | cde | 0.29 | bcd |
| 13 | | EXP-AD | 14 fl oz | @ | @ | @ | 22.8 | cde | 0.24 | bcd |
| 14 | | EXP-AF | 7 fl oz | @ | @ | @ | 24.9 | cde | 0.27 | bcd |
| 15 | Rotation | Fontelis + Teb | 20 fl oz + 8 oz | @ | --- | --- | 29.6 | cde | 0.33 | bcd |
| | | Quash | 2 oz | --- | @ | --- | | | | |
| | | Ph-D | 6.2 oz | --- | --- | @ | | | | |

* - Treatments were applied using an air-blast sprayer at a rate of 100 gal/A, all in combination with DynAmic (8 fl oz/A).

** - For evaluation of disease, a severity scale was used on 25-35 leaves of each single-tree replication: 0 (healthy), 1 (<33%), 2 (33-66%), and 4 (>66%) of leaf surface diseased.

**4. Establish baseline sensitivities, monitor for fungicide sensitivity shifts in the pathogens, and characterize mechanisms for resistance against SDHI and DMI fungicides.** DMI fungicides (FC 3) including the new Cevya were highly active against *Alternaria* isolates and against isolates molecularly identified as *V. carpophila*. A relatively narrow range of sensitivity was observed in baseline studies with DMI fungicides for *Alternaria* spp. Some outliers were detected, however, DMI fungicides remain highly effective in managing both diseases in the field.

Among FC 7 fungicides, boscalid, fluopyram, fluxapyroxad, isofetamid, and penthiopyrad are registered on almond, and pydiflumetofen, pyraziflumid, fluindapyr, and the experimental GWN10570 are in development. Baselines (i.e., for new FC 7 subgroups) or current distributions (i.e., for FC 7 subgroups already in commercial use) were established for *Alternaria* spp. for 6 SDHI fungicides (**Fig. 2**). Boscalid, pyraziflumid, and fluxapyroxad showed strong bimodal sensitivity distributions that included resistant isolates with $EC_{50}$ values of >10 ppm. A bimodal distribution was also found for fluopyram, but less sensitive isolates had $EC_{50}$ values of $\leq$0.5 ppm. Values for isofetamid and pydiflumetofen were more normally distributed and were mostly <1 or 0.07 ppm, respectively. Thus, shifts in sensitivity of *Alternaria* spp. were visualized for registered fungicides and resistance to fungicides belonging to newer SDHI sub-groups (e.g., pyraziflumid, fluxapyroxad, isofetamid) was detected in isolates that were collected before their commercial introduction. Therefore, variants most likely pre-exist, and non-detrimental mutations occur without significant fitness penalties to the pathogen. Cross resistance patterns among SDHI fungicides were evaluated in regression analyses. Relatively strong cross resistance ($R^2$=0.616) was present between fluopyram and isofetamid, weak cross resistance ($R^2$=0.406-0.416) between fluopyram and boscalid or pyraziflumid and between boscalid and pyraziflumid, and poor cross resistance ($R^2$<0.4) for the remaining fungicide pairs (**Fig. 3**).

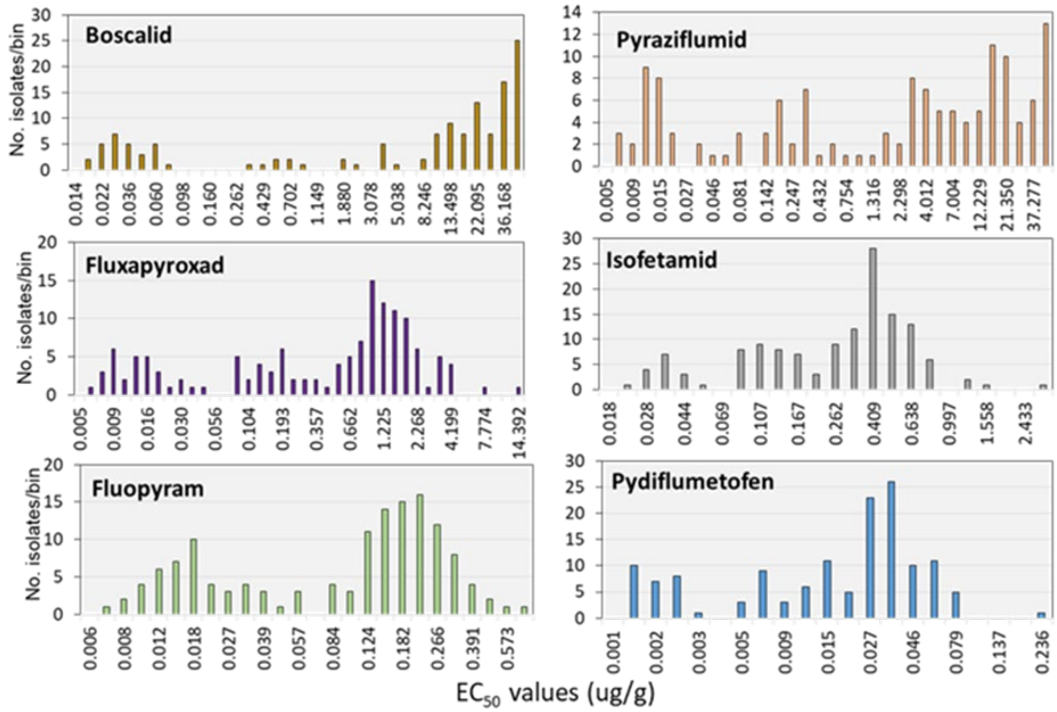
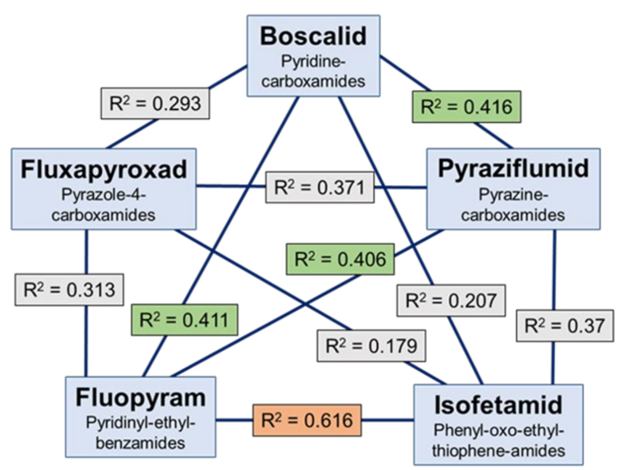



**Fig 2.** $EC_{50}$ frequency distributions of 141 isolates of *Alternaria* spp. from almond collected between 2007 and 2018 showing their sensitivity to six SDHI fungicides. Bin width h was calculated using Scott's formula: $h = 3.49sn^{-1/3}$ (Scott 1979), where s = estimate of the standard deviation and n = number of isolates.

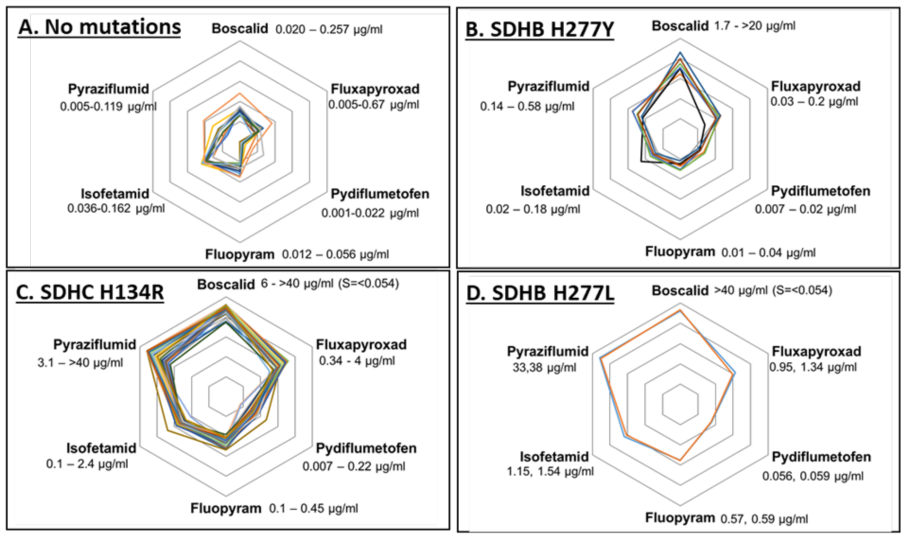
**Fig. 3.** Cross-resistance among five SDHI fungicides in 141 *Alternaria* spp. isolates from almond based on pairwise regressions of $EC_{50}$ values. $R_2$ values of >0.6 (orange), 0.4 to 0.6 (green), and <0.4 (gray) indicate a relatively strong, weak, and poor positive correlation, respectively.



Isolates of *Alternaria* spp. were used in genetic studies to correlate resistance to selected FC 7 fungicides with mutations in subunits B and C of the target SDH gene. Mutations SDHB-H277Y, SDHB-277L, and SDHC-H134R were most common, and their resistance phenotypes are shown for six SDHI fungicides in **Fig. 4**. No mutations were detected in subunit D. Mutation SDHB-H277Y that mostly targeted boscalid was determined in isolates collected after boscalid was introduced but before other FG 7 fungicides became available (Fig. 4B). With the introduction of fluxapyroxad and fluopyram, mutation SDHC-H134R became predominant and SDHB-277L was found at a low frequency. These new mutations have a broader spectrum, targeting mostly boscalid, pyraziflumid, and fluxapyroxad, but to some extent also isofetamid.

In summary, these studies indicate that cross-resistance is present among different sub-groups of FC 7 fungicides. Currently, isolates are mostly highly sensitive to fluopyram and pydiflumetofen. These should be used in mixtures or rotations with other FCs such as 3, 12, and 19 to prevent further selection of new mutations that may target these latter two SDHI fungicides.

**Fig. 4.** Fungicide sensitivity phenotypes of **A,** 15 isolates with no mutation, **B,** 7 isolates with mutation B-H277Y, **C,** 46 isolates with mutation C-H134R, and **D,** 2 isolates with mutation B-H277L. $EC_{50}$ values for each fungicide are on a $\log_{10}$ scale with 50 µg/ml at the edge of each diagram. The range of $EC_{50}$ values for isolates with each mutation is indicated.




## D. Outreach Activities – oral presentations given in 2019 and 2020

1. Jan. 9, 2019: Managing diseases in almonds. Bayer CropScience 2019 Tree, Nut, and Vine Meeting, Universal Studies, Universal City, CA. 150 people. Mainly PCAs.
2. Jan. 17, 2019: Almond Disease Management - Colusa Co. Williams, CA. 35 people. Growers and PCAs.
3. Jan. 18-19, 2019: Diseases - Key economic pests, identification, biology, and treatments in almond. Independent PCA Symposium. Monterey, CA. 65 people. Mainly PCAs.
4. Jan. 21, 2019: IPM of Almond Diseases. Syngenta Crop Protection, Bakersfield CA Pre-recorded. 45 people. Growers and PCAs.
5. Jan. 29, 2019: Almond flower, foliar, and fruit diseases and their management- Spring time and Summer diseases. Cortez, CA. 45 people. Growers and PCAs.
6. Feb. 5, 2019: Bloom and foliar diseases. UCCE Annual Almond Production Meeting, Woodland, CA. 45 people. Growers and PCAs.
7. June 25, 2019: Epidemiology and management of almond hull rot. Tulare International Agri-Center, Tulare, CA. 50 people. Growers and PCAs.
8. Nov. 5-7, 2019. Almond diseases and their management. 2019 UC Almond Short Course. Visalia Convention Center, Visalia, CA. 100 people. Growers and PCAs. State and International audience.

9. Jan. 17, 2020: Key economic pests, identification, biology, and treatments in almond. Bayer Crop Science Annual Almond Disease Management Meeting, Monterey, CA. 70 people. Growers and PCAs.
10. Jan. 21, 2020: Almond foliar diseases. North Valley Nut Conference, Orland, CA. 250 people. Growers and PCAs as well as regulators and industry representatives.
11. Jan. 22, 2020: Fungal and bacterial almond disease management during bloom. UCCE Colusa Winter Almond Meeting, Williams, CA. 35 people. Growers and PCAs.
12. Jan 29, 2020: Key economic diseases, identification, biology, and treatments in almond. Bayer Crop Science Chico Tree Nut Meeting, Chico, CA. 250 people. Growers and PCAs as well as regulators and industry representatives.
13. Feb. 5, 2020: Almond flower, foliar, and fruit diseases and their management. Yolo-Solano-Sacramento Almond Meeting, 50 people. Growers and PCAs.

**E. Materials and Methods:**

***I. Etiology - Determine the* V. carpophila *population composition within selected orchards and determine if sexual reproduction occurs within orchard populations.***
Previously, AFLP data and Parsimony Tree Length Permutation Tests of three populations of *V. carpophila* were used to test the hypothesis that sexual recombination is occurring. RAPD and UP-PCR genotyping, UPGMA bootstrap analysis, and DNA microsatellites were used to determine diversity among isolates from different hosts, including almond. Growth rates were determined at temperatures from 10 C to 35 C. Conidial germination was evaluated on PDA for initial viability, or suspensions were placed on glass slides and air-dried at 25 C, 24% RH. Spores were re-hydrated on days 3, 4, 5, and 7 and spore germination was determined on PDA. Percent conidial germination was ln-log transformed and regressed over time. Equations for the regressions and $R^2$ values were determined.

***II. Management***

**A. Dormant treatments for scab management**. Bravo-, captan-, and ziram-oil (3% summer spray oil) were evaluated for scab management. Applications were made in January using an air-blast sprayer calibrated for 100 gal/A using labeled rates. Twig lesions were monitored in the spring, and the onset of sporulation was determined. Incidence and severity of scab on fruit were evaluated and in-season applications were made at the onset of sporulation. Data were analyzed using analysis of variance and multiple mean separation methods of SAS.

**B.** ***Evaluate new fungicides and mixture/rotation programs for their efficacy in managing scab and Alternaria leaf spot.*** Fungicides and biologicals including natural products that were evaluated are shown in Tables 1-4. Fungicides were used as single-fungicide, mixtures, and rotation programs. Applications were done in commercial orchards with a history of disease. Applications for Alternaria spot were done based on the modified DSV model approximately in mid-April, early-May, and mid-May. In-season applications for scab were initiated as described above.

Alternaria leaf spot was evaluated based on incidence and severity of infected leaves as well as tree defoliation, and fruit were evaluated for scab using four single-tree replications. Rating scales are indicated in the footnotes of each table. Incidence and severity of scab on almond fruit were evaluated in mid/late summer. Other diseases such as rust or leaf blight were evaluated if present.

**C.** ***Establish baseline sensitivities and monitor shifts in sensitivity of* Alternaria *and* Fusicladium *spp. against SDHI fungicides and characterize resistance mechanisms for***

***SDHI and DMI fungicides.*** Using the SGD method, in vitro sensitivities of isolates from several locations were determined for mefentrifluconazole (Cevya) and SDHIs shown in Fig. 2. Sequence analyses of *sdh* gene sub-units *B, C,* and *D* were performed for isolates of *Alternaria* spp. with different resistance levels to SDHI sub-groups. Published primers were used for amplification of the *sdh* sub-units. Cross-resistance among five SDHI fungicides in 141 isolates was based on pairwise regressions of $EC_{50}$ values. For *V. carpophila,* primers for the *cyp51* gene were developed based on multiple sequence comparisons from other fungi to determine the mechanism of DMI resistance. Sequencing reactions were done using standard protocols. The partial sequence of the *cyp51* gene was expanded, and sequences were compared among sensitive and resistant isolates to determine the occurrence mutations.

## F. Publications that emerged from this work

### Refereed Journal Articles -


1) Adaskaveg, J. E., and Förster, H. 2015. Dormant treatments with chlorothalonil-oil delay the production of primary inoculum of almond scab caused by *Fusicladium carpophilum*. Phytopathology 105(Suppl. 4):S4.3.
2) Adaskaveg, J. E., Luo, Y., and Förster, H. 2020. Characterization of resistance to five SDHI sub-groups in *Alternaria* species causing leaf spot of almond in California. Pages 173-180, in: Modern Fungicides and Antifungal Compounds. Vol. IX. H. B. Deising, B. Fraaije, A. Mehl, E. C. Oerke, H. Sierotzki, and G. Stammler, eds., Deutsche Phytomedizinische Gesellschaft, Braunschweig, Germany. https://plant-protection.net/fileadmin/documents/Verlag/02_SP/05_Reinhard/0294-sp-2020-reinh-8.pdf
3) Bock, C., Young, C., Zhang, M., Chen, C., Brannen, P. M., Adaskaveg, J. E., and Charlton, N. 2021. Mating type idiomorphs, heterothallism and high genetic diversity in *Venturia carpophila*, cause of peach scab. Phytopathology. *In Press.* https://doi.org/10.1094/PHYTO-12-19-0485-R
4) Chen, C., Bock, C.H., Brannen, P.M., Adaskaveg, J.E., Hotchkiss, M.W., Brewer, M., and Wood, B.W. 2014. Characteristics of genetic variability among isolates of *Fusicladium* species from peach, almond and pecan in the USA. Phytopathology 104:Suppl. 3 S3.16.
5) Chen, C., Bock, C. H., Brannen, P. M., and Adaskaveg, J. E. 2018. Mining and characterization of microsatellites from a genome of *Venturia carpophila*. Mycological Progress 17:885–895. https://doi.org/10.1007/s11557-018-1401-x


### Other publications (e.g., outreach materials) –


1) Adaskaveg, J. E., and Michailides, T. J. 2021. Efficacy and Timing of Fungicides, Bactericides, and Biologicals for Deciduous Tree Fruit, Nut, Strawberry, and Vine Crops (PDF). *In production*. http://ipm.ucanr.edu/PDF/PMG/fungicideefficacytiming.pdf
2) Ziram: Re-Registration Eligibility Decision Review. Support letter posted on EPA website. 2020-21. https://www.regulations.gov/docket?D=EPA-HQ-OPP-2015-0568
3) Prepared several disease chapters for the Almond Production Manual.
4) Haviland, D. R., Symmes, E. J., Adaskaveg, J. E., Duncan, R. A., Roncoroni, J. A., Gubler, W. D., Hanson, B., Hembree, K. J., Holtz, B. A., Stapleton, J. J., Tollerup, K. E., Trouillas, F. P., and Zalom, F. G. Revised continuously. *UC IPM Pest Management Guidelines: Almond*. UC ANR Publication 3431. Oakland, CA. https://www2.ipm.ucanr.edu/agriculture/almond/